GEOCHEMISTRY AND MICROBIOLOGY OF GEOTHERMAL AEROSOLS IN ICELAND: IMPLICATIONS FOR BIOSIGNATURES IN THE PLUMES OF ENCELADUS. M. G. Fox-Powell[1], B. Stephens[1], C. Batty[1], T. Gladding[2] and K. Olsson-Francis[1], [1]AstrobiologyOU, Open University, Milton Keynes, UK (mark.fox-powell@open.ac.uk), [2]School of Engineering & Innovation, Open University, Milton Keynes, UK.

Introduction: The plumes of Enceladus offer a unique opportunity to determine if life currently exists within the moon's likely habitable subsurface ocean [1]. Cassini's encounters with the plumes have implied that plume ice grains originate as aerosolized ocean fluid, produced by vigorous bubbling of hydrothermal gases at the liquid-vapor interface [2]. Seawater aerosols on Earth can be rich repositories of microorganisms [3] and biological molecules [4], raising the possibility that if life exists at Enceladus, plume particles originating as aerosols may also contain biosignatures. However, seawater aerosol formation involves the breaking of waves and the presence of dense populations of photosynthetic organisms; conditions drastically different from those expected at Enceladus [3].

We investigated the chemistry and microbial content of aerosol plumes originating from actively bubbling geothermal springs in Iceland, to better understand the potential for biosignature entrainment in the plumes of Enceladus. Icelandic geothermal springs provide excellent natural laboratories for studying plume aerosols, with aerosolization driven by bubbling of hydrothermal gases analogous to those likely driving plume formation at Enceladus [2], anoxic fluids with chemistries relevant to Enceladus and other icy moons [5], and microbial communities adapted to these chemical conditions [6]. In contrast to plume aerosols at Enceladus, geothermal aerosols on Earth emerge from accessible fluid reservoirs, offering the opportunity to fully interpret biosignatures in the context of known biological activity that produced them.

Field localities: Two locations in south-west Iceland were sampled in August 2022: the Strokkur geyser at Geysir (N64.312712 W020.300761), and a system of three bubbling springs at Ölkelduháls (N64.056677 W021.235075) (Fig. 1A). These locations were selected to encompass contrasting aerosolization regimes: periodically erupting geyser plumes (Strokkur) vs constantly bubbling springs (Ölkelduháls). Strokkur is a silica-rich spring that discharges geyser eruptions reaching 10-30 m height approximately every 5-8 minutes (Fig. 1B). The temperature of the Strokkur spring, close to the geyser origin, was measured to be 73.5 °C with a pH of 8.7. The Ölkelduháls system is sulfur-rich and comprises three pools closely situated (Fig. 1C), each of which experiences constant bubbling. Pools exhibited temperatures between 59.3 and 73.6 °C and pH levels between 3.4 and 6.5.

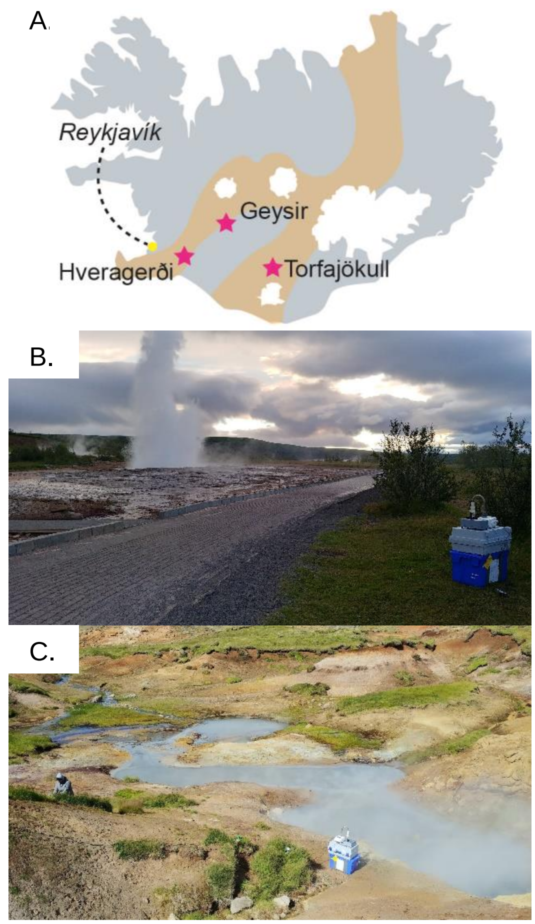

Figure 1. A: Map of Iceland showing sampling locations (stars), volcanic rifting zone (orange), and glaciers (white); B: Strokkur geyser eruption; C: Ölkelduháls springs.

Methods: *Sampling.* Aerosols were sampled in 30-minute increments using a Bertin Instruments Coriolis μ aerosol sampler at 300 liters air min^{-1}. Aerosol flux was monitored across three size fractions (diameters ≤ 10.0, ≤ 2.5 and ≤ 1.0 μm) once per second during sampling intervals using a Turnkey Osiris Particle Enumerator. Replicate samples were collected for microbiological and geochemical analyses of the aerosol phase. One set of upwind background samples were obtained at each site, and sampling locations downwind were guided by aerosol fluxes recorded by the Osiris. Samples of spring fluids were also taken for geochemical and microbiological analyses, and

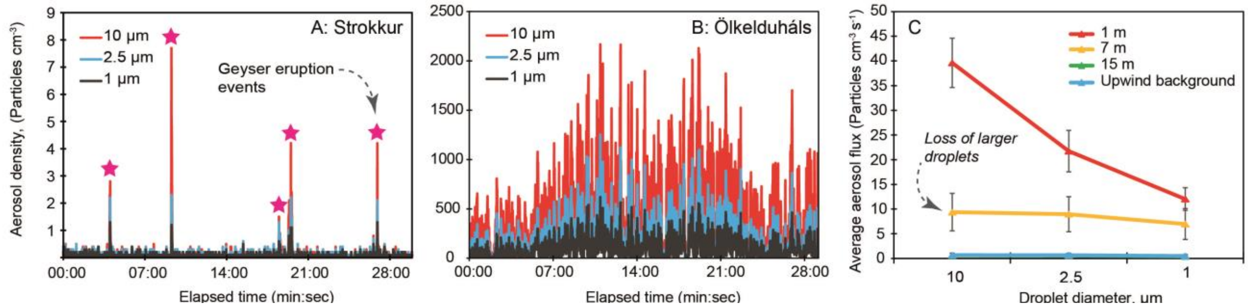

Figure 2. A, B: Aerosol fluxes measured over 30 min. intervals at Icelandic field sites. C: Average aerosol fluxes recorded over 30 min. intervals at increasing distances from an Ölkelduháls bubbling hot spring. Bars indicate ± 1 S.D.

geothermal gases responsible for bubbling were sampled using thermal desorption (TD) tubes.

Analysis. Inorganic ion concentrations and elemental composition of aerosol and spring samples were analyzed using ion chromatography, inductively coupled plasma-mass spectrometry and optical emission spectroscopy. Microbial abundances in aerosol and spring samples were determined by direct counts under fluorescence microscopy. Ongoing work includes community profiling of microorganisms in aerosol and spring samples using 16S rRNA amplicon sequencing, gas profiling with gas chromatography-mass spectrometry, and analysis of solid mineral particulates in aerosol samples with scanning electron microscopy-energy dispersive spectroscopy.

Results and Discussion: Aerosol flux monitoring revealed aerosol densities downwind of geothermal springs 2-4 orders of magnitude higher than background levels (Fig. 2), demonstrating that bubbling geothermal springs are prolific local sources of aerosols. We also found that aerosol fluxes were tightly controlled by bubbling and/or geyser eruption activity: Strokkur fluxes were directly, and strongly, influenced by geyser eruptions, as shown in Fig. 1b. Immediately following eruption events, aerosol densities momentarily spiked before rapidly falling to background levels as the aerosol plume dispersed. By contrast, Ölkelduháls, which exhibited much less vigorous, but sustained, bubbling produced high densities of aerosols throughout sampling periods. These findings illustrate the importance of bubbling dynamics for defining aerosol flux, thus constraining the nature and rate of aerosolization at Enceladus will be vital to understanding the origins of the plumes.

Preliminary cell enumerations show that microbial abundances within aerosols in the immediate vicinity of springs were elevated 2-3 orders of magnitude above background, indicating that microbial biomass can be ejected by bubbling of hydrothermal gas. However, aerosol size distributions at Ölkelduháls were influenced by distance from the spring, with the largest measured droplets (≤ 10.0 µm diameter) decreasing to background levels within 10 m. If biomass tends to associate with the largest droplets, it may quickly fall out and therefore not be dispersed. Because the Enceladus plumes are stratified by size, [7], understanding size-dependent variations in aerosol biomass content will lead to better predictions of the distribution of potential biosignatures in the plumes, and the composition of ice-grains ejected to the altitude of spacecraft fly-throughs.

Ongoing work: Our ongoing work is applying molecular tools to determine whether the microbial assemblages in geothermal aerosols represent the full diversity of source spring communities, or whether aspects of the spring communities are ejected more efficiently than others. Combining microbiological data with geochemical data will allow us to quantitatively compare a given aerosol sample to the aerosol source, establishing the extent to which biogeochemistry can be faithfully reconstructed from the aerosol sample and how this varies with distance.

Our findings provide the first insights into the potential expression of biosignatures in cryovolcanic plumes at Enceladus. Findings will enable new perspectives on Cassini plume fly-through data and can be leveraged to predict plume behavior and composition at other planetary bodies with possible plumes, such as Europa.

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